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Pine Nuts: A Review of Recent Sanitary Conditions and Market Development

Hafiz Umair Masood Awan ¹  and Davide Pettenella ^{2,*} 

¹ Department of Forest Science, University of Helsinki, FI 00014 Helsinki, Finland; awan48@gmail.com or hafiz.awan@helsinki.fi

² Department Land, Environment, Agriculture and Forestry, University of Padova, 35020 Legnaro (PD), Italy

* Correspondence: davide.pettenella@unipd.it; Tel.: +39-049-827-2741

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Abstract: Pine nuts are non-wood forest products (NWFP) with a constantly growing market notwithstanding a series of phytosanitary issues and related trade problems. The aim of this paper is to review the literature on the relationship between phytosanitary problems and trade development. Production and trade of pine nuts in Mediterranean Europe have been negatively affected by the spreading of *Diplodia sapinea* (a fungus) associated with an adventive insect *Leptoglossus occidentalis* (fungal vector), with impacts on forest management, production and profitability and thus in value chain organization. Reduced availability of domestic production in markets with a growing demand has stimulated the import of pine nuts. China has become a leading exporter of pine nuts, but its export is affected by a symptom caused by the nuts of some pine species: ‘pine nut syndrome’ (PNS). Most of the studies mentioned in the literature review concern PNS occurrence associated with the nuts of *Pinus armandii*. We highlight the need for a comprehensive and interdisciplinary approach to the analysis of the pine nuts value chain organization, where research on food properties and clinical toxicology may be connected to breeding and forest management, forest pathology and entomology, and trade development.

Keywords: pine nut; pine nut syndrome (PNS); pine mouth syndrome (PMS); cacogeusia; non-wood forest products; *Leptoglossus occidentalis* (Western conifer seed bug); *Diplodia sapinea* (*Sphaeropsis blight*)

1. Introduction

There is a great variety of species in the genus *Pinus* that produce nuts of commercial interest. This variety is connected to tree productivity, nutritional and medicinal value, taste and textural properties, and commercial value of the nuts. Despite these variations, when retailed, they are all included under the generic name ‘pine nuts’ [1].

This paper reviews the recent literature on pine nuts from an interdisciplinary perspective. The motivations for our survey are due to the remarkable development of the international pine nuts market; a development based more on empirical evidence than on systematic and clear results deriving from production and trade data recorded by statistical institutions. For these reasons, we try to highlight the growing role played by this category of products in the global market, with a focus on the European Union (EU) market which has seen the most radical changes in its structure, and the main issues considered by the scientific literature, i.e., pine forest phytosanitary problems and human health problems connected to the consumption of some species of pine nuts.

The paper is structured as follows: the introductory section highlights the historical origins of pines and some general aspects of the pine nut sector, such as the nutritious and dietary values. Details of the research methodology in the description of market development and literature review follow. We then present the results of this combined analysis: the production and trade patterns at the

world and EU level and the current issues that hamper market development, making reference to pine nut syndrome (PNS), pathogenic fungi like *Diplodia sapinea* (Fr.) Fuckel [2,3] or *Sphaeropsis blight* and the related pest *Leptoglossus occidentalis* Heidemann (the western conifer seed bug). Lastly, conclusions and suggestions are made for further research.

2. Pines and Pine Nuts: General Aspects

It is thought that the first pines emerged somewhere in northern Asia around 180 million years ago. Geologically, it was the Triassic Period in the Mesozoic Era (age of reptiles) [4–6]. Pines gradually differentiated into Haploxylon (soft or white) pines and Diploxylon (hard or yellow) pines [7] (pp. 51–52) about 75 million years ago [4]. Today, these two distinct sub-genera are still considered to be in the world's flora classification [4] (see [4,6] for further historical and cultural details). They are both important sources of seeds for human consumption. Humans started to utilize pine nuts in the Paleolithic era [8]; the Food and Agriculture Organization reported the utilization of pine nuts as a comestible food item starting from pre-historic times [9,10]. Today's busy lifestyles provide an opportunity to use tree nuts as they are nutritious, handy and tasty snacks, and pine nuts are used in traditional recipes in many countries.

The pine nut species that are consumed domestically may differ from those that have an important commercial role and are traded [11,12]. For instance, *Pinus armandii* Franch. (Chinese white pine), *Pinus tabulaeformis* Carr (Chinese red pine), *Pinus yunnanensis* Franch (Yunnan Pine), and *Pinus massoniana* Lamb (Masson's pine) are mainly imported into Europe from China and differ from the domestically produced *Pinus pinea* L. nuts [13] (p. 2), [14]. A reliable online working list of all plant species currently contains 636 scientific species records for the genus *Pinus* [15]. Of these 636 records, 126 are accepted species names. Of these 126 species of *Pinus* that exist worldwide, only 29 provide edible nuts according to the Food and Agriculture Organization [9], while 20 are traded locally or internationally. The species that are commercially important for providing pine nuts are listed in Table 1.

Table 1. Comestible species of pine nuts in the world.

Species	Natural Range	Remarks
Soft Pines (Haploxylon)		
<i>P. ayacahuite</i> Engelm. ex Schlftl.	Central America, Mexico	Traditional food for indigenous tribes
<i>P. albicaulis</i> Engelm	United States and Western Canada	Traditional food for indigenous tribes
<i>P. cembra</i> L.	Europe (Alps and Carpathian Mountains)	Locally important
<i>P. flexilis</i> E.James	United States and Western Canada	Traditional food for indigenous tribes
<i>P. gerardiana</i> Wall. ex D.Don	Pakistan, East Afghanistan, North India	Important in international trade
<i>P. koraiensis</i> Siebold & Zucc.	East China, Japan, Korea, South-east Siberia	Important in international trade
<i>P. lambertiana</i> Douglas	Western United States (California, Oregon)	Traditional food for indigenous tribes
<i>P. monticola</i> Douglas ex D.Don	North-west United States and adjoining Canada	Traditional food for indigenous tribes
<i>P. pumila</i> (Pall.) Regel	East Siberia, East China, Japan, Korea	Locally important
<i>P. sibirica</i> Du Tour	Mongolia, Russia (Central Siberia)	Nuts are ground into cooking oil
<i>P. strobiformis</i> Engelm	North Mexico, South-west United States	Traditional food for indigenous tribes
Pinon pines	North Mexico, South-west United States	A complex group of about 13 species. Many are important food sources
Hard Pines (Diploxylon)		
<i>P. coulteri</i> D.Don	California (United States)	Traditional food for indigenous tribes
<i>P. pinea</i> L.	Mediterranean Europe and Near East	Important in international trade
<i>P. ponderosa</i> Douglas ex C.Lawson	United States and Western Canada	Traditional food for indigenous tribes
<i>P. sabiniana</i> Douglas ex D.Don	California (United States)	Traditional food for indigenous tribes
<i>P. roxburghii</i> Sarg.	India, Pakistan	Traditional food source
<i>P. torreyana</i> Parry ex Carr.	California (United States)	Traditional food for indigenous tribes

Source: FAO [16] and Ciesla [9].

Of these 20 species, only three are of much international significance: *P. koraiensis* from China, *P. pinea* from southern Europe, *P. gerardiana* from Pakistan and Afghanistan [16,17]. Handy et al. [18] mentioned *Pinus edulis* Engelm, *P. monophylla* Torr and Frém, *P. pumila* (Pallas) Regel, *P. sibirica*,

P. wallichiana A. B. Jacks, *P. tabuliformis* Carr., *P. yunnanensis* Franch., *P. gerardiana*, *P. koraiensis*, *P. massoniana* Lamb, as the most relevant species exported worldwide. Other species are usually sold in local markets where they originate [10].

Pine nuts are popular in different parts of the world, especially in the Mediterranean, Middle East, Asia, and in the southwestern United States by Native Americans [19] (p. 285). The majority of ecological and silvicultural research has been focused on *P. pinea* [8,20–22]. It has been stated that in some rain-fed areas (i.e., Mediterranean areas) gathering of cones containing pine nuts yields a much better income in a short period than harvesting wood after long rotations [22].

Tree nuts can be defined as “dry fruits with generally one seed in which the overall wall becomes hard at maturity” [23]. However, a pine nut is not a true nut because it lacks an outer carpel (though it is a tree nut). They are instead edible small-sized ivory-coloured nutritious seeds [10,17,19,24], which are collected from the cones of different pine species [24,25] and are oil rich [13].

The kernels of European stone pine (*P. pinea*), which is indigenous to the Mediterranean area, are sometimes described as Mediterranean pine [26] or Italian pine nuts. In Neo-Latin languages, pine nuts are known as *pinoli* or *piñone* (in the US they are often called *pignoli*). In the other parts of the world, kernels of the pinyon pines (*P. edulis* and *P. monophylla*), which grow in the southwestern US and in northern Mexico, are known as pinon nuts [25].

Pine nuts are a source of valuable nutrients (Table 2). *P. pinea* supplies double the amount of protein than Chinese (*P. koraiensis*) and Pakistani pine nuts (*P. gerardiana*) [27]. However, the latter two species provide more carbohydrate and fats, which are very important components of our diet. Pakistani pine nuts in particular are a potentially rich source of the carbohydrates that may be the most useful for athletes.

Table 2. Nutrient values and weights for edible portion of pine nuts.

Nutrient	Unit	Value Per 100 g	1 Cup (135 g)	1 oz (167 Kernels) or (28.35 g)	10 Nuts (1.7 g)
Proximates					
Water	g	2.28	3.08	0.65	0.04
Energy	kcal	673	909	191	11
Protein	g	13.69	18.48	3.88	0.23
Total lipids (fat)	g	68.37	92.3	19.38	1.16
Carbohydrate, by difference	g	13.08	17.66	3.71	0.22
Fiber, total dietary	g	3.7	5	1	0.1
Sugars, total	g	3.59	4.85	1.02	0.06
Minerals					
Calcium, Ca	mg	16	22	5	0
Iron, Fe	mg	5.53	7.47	1.57	0.09
Magnesium, Mg	mg	251	339	71	4
Phosphorus, P	mg	575	776	163	10
Potassium, K	mg	597	806	169	10
Sodium, Na	mg	2	3	1	0
Zinc, Zn	mg	6.45	8.71	1.83	0.11
Vitamins					
Vitamin C, total ascorbic acid	mg	0.8	1.1	0.2	0
Thiamin	mg	0.364	0.491	0.103	0.006
Riboflavin	mg	0.227	0.306	0.064	0.004
Niacin	mg	4.387	5.922	1.244	0.075
Vitamin B-6	mg	0.094	0.127	0.027	0.002
Folate, DFE	µg	34	46	10	1
Vitamin B-12	µg	0	0	0	0
Vitamin A, RAE	µg	1	1	0	0
Vitamin A, IU	IU	29	39	8	0
Vitamin E (alpha-tocopherol)	mg	9.33	12.6	2.65	0.16
Vitamin D (D2 + D3)	µg	0	0	0	0
Vitamin D	IU	0	0	0	0

Source: United States Department of Agriculture (USDA), National Nutrient Database [28].

Table 2. Cont.

Nutrient	Unit	Value Per 100 g	1 Cup (135 g)	1 oz (167 Kernels) or (28.35 g)	10 Nuts (1.7 g)
Vitamin K (phyloquinone)	µg	53.9	72.8	15.3	0.9
Lipids					
Fatty acids, total saturated	g	4.899	6.614	1.389	0.083
Fatty acids, total monounsaturated	g	18.764	25.331	5.32	0.319
Fatty acids, total polyunsaturated	g	34.071	45.996	9.659	0.579
Cholesterol	mg	0	0	0	0
Other					
Caffeine	mg	0	0	0	0

Source: United States Department of Agriculture (USDA), National Nutrient Database [28].

Pine nuts are beneficial for checking blood lipids and controlling coronary heart disease (CHD) [29]. This is due to their containing only unsaturated fatty acids, whereas most other nuts also have monounsaturated fatty acids, primarily oleic acid. However, pine kernels contain mostly linoleic acid in the form of polyunsaturated fatty acids. Linoleic acid can be transformed into cellular mediators that play an important role at the vessel level and improve blood coagulation [30]. *P. koraensis* Siebold and Zucc and *P. sibirica* have the highest fat content—maximum 65–75%. Apart from their favorable association with cardiovascular health, pine nuts are a rich source of other nutrients [31]. Table 3 presents a comparison of dietary values among species available in the world market. Interestingly, pine nuts may also be used as natural appetite suppressants [19,32,33].

Table 3. Dietary values of several species of pine nuts in comparison with other commercially important nuts ¹.

Type of Nut	Carbohydrates (%)	Fats (%)	Protein (%)
<i>P. pinea</i>	7	48	34
<i>P. gerardiana</i>	23	51	14
<i>P. edulis</i>	18	62–71	14
<i>P. sibirica</i>	12	51–75	19
<i>P. cembroides</i> Zucc.	14	60	19
<i>P. monophylla</i>	54	23	10
<i>P. koraiensis</i>	12	65	18
<i>P. sabiniana</i>	9	56	28

¹ Percentages are approximate and based on shelled nuts. Source: Evaristo et al. [34]; Mutke et al. [27].

The chemical contents of pine nuts vary according to two main factors i.e., geographical range and climate [35,36]. European pines (*P. pinea*) have specific fatty acids profiles that differ from Pakistani pine nuts (*P. gerardiana*). This aspect has relevant interest concerning the issue of PNS as discussed in the section on the results of the literature review.

3. Materials and Methods

The study is based on two connected analyses and related methodologies: A market survey to provide robust evidence on the increased importance of the international pine nut trade, and a literature review to highlight the underlying problems connected with the expansion of the nut market—the phytosanitary aspects affecting pine forests and human health issues connected with nut consumption.

EUROSTAT provided information for the elaboration of export and import data. The product code (08029050 Pine "*Pinus* spp." nuts, fresh or dried, whether shelled or not, or peeled) in the database for international trade since 1988 (CN8 DS-016890) was used. However, we set 2006 as the base year to avoid working with missing data prior to that year. From the EUROSTAT database average unit values (€/Kg) and their annual variation have been elaborated for the five key EU trade players and

for the 28 EU member states as an aggregate (EU28). EUROSTAT trade data have been expressed as extra-EU (for transactions with all countries outside of the EU) and intra-EU (all transactions occurring within the EU).

For the literature review, we mainly used the Web of Science (WoS); the database was accessed during January 2016–May 2017 and we did not limit our search to specific years, even if the main publications considered are those of the last three decades.

4. Results

4.1. Production and Trade

Pine nuts are not commercially exported from Oceania [17]. They are mainly imported from China, Spain, Italy and Turkey [17]. China and Pakistan have become main exporters over the past decade to meet world demand [8,12].

The most popular edible variety is the European stone pine or umbrella pine (*P. pinea*), which has been consumed by humans for more than 20 centuries [26,37]. Today, the Ligurian pesto from Italy has pine nuts as an ingredient [38].

In recent years, market growth has been observed particularly in the US, becoming a 100 million US\$ market there [10,29]. The demand for pine nuts by consumers in the US has increased for the last 10 years [18]. This trend might be explained by a recent Irish study that gave evidence that nut consumption can help in decreasing the risk of coronary heart disease (CHD) and sudden cardiac arrest [29]. There has recently been a hike in pine nut prices as gourmet products [10]. For instance, in The Netherlands, 100 g of pine nuts costs 4 € [10] while in Finland it can cost up to 5–6 € depending on the retailer. Three species of pine kernels (*P. gerardiana*, *P. koraiensis*, and *P. pinea*) as described in FAO [16] have been part of global trade for many years, whereas *P. sibirica* entered the international market in 2002 [39].

Tracking pine nuts along the supply chain is of increasing importance. Cases where nut species were erroneously labeled due to lack of origin information have frequently been reported. Moreover, re-exports among countries hinder the traceability of product origin. It occurs quite often that a consumer is not supplied with adequate and suitable information regarding geographical origin, or even the species of imported pine nuts on a product label.

Data are not available on world production of pine nuts [16]. The species are quite difficult to distinguish from one another. According to Mutke et al. [27], even EUROSTAT and the Codex Alimentarius have not devised differentiation procedures for pine nut species. Also in the case of trade analysis the lack of specific trade codes (e.g., 0802.90.50 code in EUROSTAT database for pine nuts versus 0802.90 in the UN COMTRADE database) limits the tracing of a gourmet product like *P. pinea* nuts that yields a wholesale price of 45,000 to 60,000 €/t in Europe.

T and opportunities for policy makers” [40]. The report clearly states that pine nuts could not be included in the two main nut categories proposed (the first consisted of coconuts, Brazil nuts and cashew nuts, while the second gathered all the others—hazelnuts, walnuts, chestnuts and pistachios—excluding pine nuts). According to Vidale et al. [40], the main obstacle for the inclusion of pine nuts in the analyses was the single code used for mixed commodities of chestnuts, pistachios and pine nuts together (reference HS6 code of the Multipurpose International Product Nomenclature developed by the World Customs Organization).

According to the global statistical review 2014–2015 published by the International Nut and Dried Fruit Council Foundation [15], world pine nut production was estimated at 39,950 MT (kernel basis). This was 86% higher than in 2004. The increase was due to the expansion of China’s production capacity which had grown 10 times since 2004. Indeed, China was the principal producer in 2014. The crop yielded 25,000 MT (62% of total production share). Korea DPR came next (5000 MT or 12% share), followed by Afghanistan (3100 MT or 8% share) and Pakistan (3000 MT or 7% share). Russian Federation contributed 2500 MT (6% share) to the world market (Geisler 2016). At the end of

2013, pine nut exports totaled 14,000 MT, i.e., 1.5 times the quantity exported in 2008. These observations seem to confirm the export recovery trend that started in 2009. China shipped 10,683 MT, 76% of total pine nut exports making it the main exporter; 29% of these exports went to the US. Note that China appeared in the U.S. Bureau of the Census as an exporting country of pine nuts for the first time in 1976 [41].

The price development in Figure 1 was estimated as average real price (2006–2016) of imported products to the EU. EU imports are dominated by five countries (which cover about 98% of the extra-EU import market in all the years considered). Prices increased at a remarkable rate, having doubled in the last 10 years.

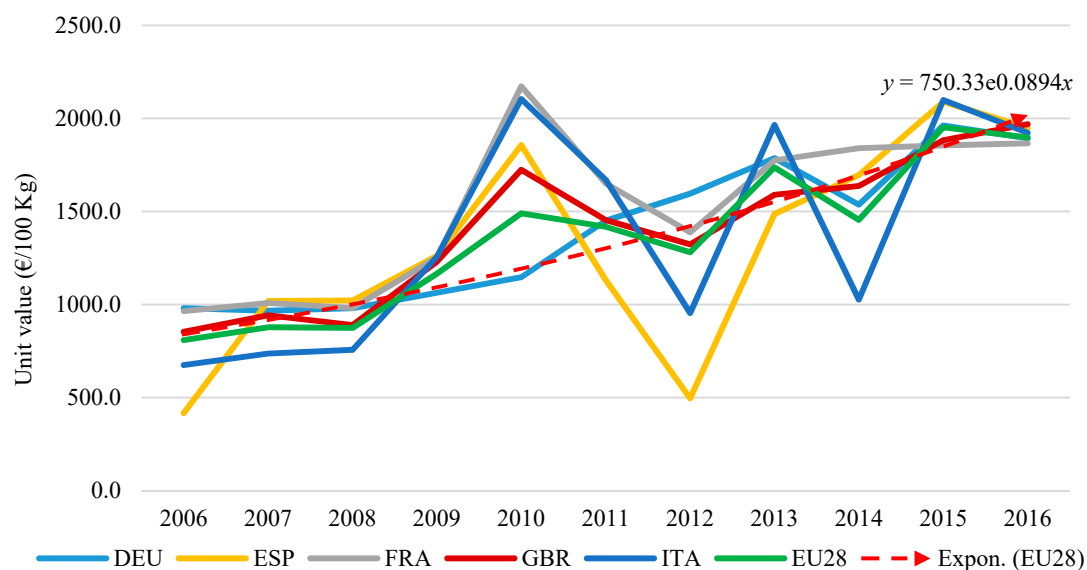


Figure 1. Price development estimated as average current prices of products imported to the EU (2006–2016).

It is evident that unit values fluctuate sharply. Unlike in Italy and Spain, unit prices did not dramatically shift in France, Germany and Great Britain. In fact, the exponential trend line increases gradually over a number of years. This is an indication that trade is developing positively in terms of demand for pine nuts in all 28 EU member states.

We have found some interesting research findings [1] concerning retailers of pine nuts all around the world. A full list of countries and retailers that sold problematic pine nuts is represented in tabular form (supplementary information). It is not the intention to ruin the reputations of the retailers nor to suggest that buyers boycott them. It is just to give an idea that development of pine nut syndrome to such a global extent is a serious issue. The names of pine species are usually missing from the packet, which makes it almost impossible to trace their botanical origin [12]. Table 4 presents the total number of PNS cases reported in 2011. It indicates that Australia, Canada, England, the Netherlands, and the USA were the top five countries for the problem out of the 22 countries surveyed in the study.

Table 4. Summary of pine nut syndrome (PNS) cases reported during 2011.

Countries	Number of Cases Reported	Reported Cases (%)
Australia	15	4
Belgium	4	1
Canada	20	5
Czech Republic	1	0
Denmark	7	2
England	44	11
Finland	1	0
Germany	3	1
Hungary	1	0
Iceland	1	0
Ireland	3	1
Malta	1	0
Netherlands	51	13
New Zealand	4	1
Norway	3	1
Poland	1	0
Scotland	2	1
Singapore	1	0
Slovenia	1	0
South Africa	4	1
Sweden	6	2
USA	221	56
Total	395	100

Source: own elaboration from Tan [1]. The number of cases above 3 (or 1%) are indicated in italics.

4.2. Current Main Issues Affecting Pine Nuts Production and Trade

4.2.1. Pine Nut Syndrome

In 2001, Mostin first introduced the term ‘Pine Nut Syndrome’ (PNS) in the literature as a taste disturbance [13,36,42]. Pine nut syndrome is also known as pine mouth syndrome (PMS) or pine nut mouth syndrome [17,24,36,43–46]. Kwegyir-Afful et al. [47] introduced the term ‘Pine Mouth Event’. Medically, it is known as dysgeusia, metallogueusia or cacogeusia [10,43,46,48].

According to Hampton et al. [48], the initial symptom is a persistent metallic or bitter taste starting within hours to the first two days after eating pine nuts and can last for up to 14 days. In exceptional cases, the metal or acid taste could last for a maximum of 42 days [48]. These pine mouth events went unnoticed by the scientific community until cases reported by pine nut consumers rose dramatically between 2008 and 2012 [27,42]. Consequently, several studies tried to investigate the taste disturbance due to ingestion of pine kernels.

Apart from health aspects, lipids in pine nuts are of special importance due to their use in identification of species causing the syndrome. Senter et al. [49], in their gas-liquid chromatography-mass spectrometry experiment, suggested that phenolic acids in pine nuts ($1.37 \mu\text{g g}^{-1}$) are relatively higher than in almonds ($1.08 \mu\text{g g}^{-1}$), filberts ($0.36 \mu\text{g g}^{-1}$) and black walnuts ($0.51 \mu\text{g g}^{-1}$). Phenolic compounds might be the potential reason for taste disturbance as these possess an astringent quality [49,50]. Weise [51] stated that the pine mouth events can happen due to rancidity of lipids. Destailats et al. [12] negated association of PNS to the oxidative reactions [36] of lipids identified in their gas liquid chromatography analyses. It was also shown by Destailats et al. [12] that the total composition of fatty acids in Chinese kernels (*P. armandii*) did not differ markedly from the other pine species in their experiment. From these studies, Köbler et al. [50] inferred that the fatty acid composition of pine nuts does not influence taste disturbance. Möller [24] proposed that PNS may be due to an elevated level of bile juice, which is produced as a result of bioactive compounds in pine

nuts. The effect might be compounded by enterohepatic recirculation, which dramatically extends the residence time of many chemicals or drugs in the digestive tract.

No toxin, fungus, pesticide component or heavy metal had been evaluated as a direct reason for generating PNS [36]. To the best of our knowledge, none of the studies (Table 5) provided a relevant single cause for PNS development, though reliable methods were used, such as gas liquid chromatography [12], fatty acid analysis and DNA testing [37], and/or nuclear magnetic resonance analysis [50] to identify pine nut species from the reference samples. Even the most extensive study on PNS conducted in France by Flesch et al. [45], proved unhelpful in this regard. Flesch et al. [45] suggested that the bitterness might be caused by an unknown toxin. The toxin seemed to them to be heat resistant. PNS is more frequent in females than males. They also hypothesized that this taste disturbance might be due to a polymorphism in the genetic expression of taste functions.

In addition to PNS, Hampton et al. [48] briefly introduced the term anaphylaxis. For further details regarding anaphylactic reactions, Cabanillas and Novak [26] have reviewed the issue quite clearly.

Most of the research in forestry dealt with pine trees (e.g., mainly ecology, historical aspects, and tree health issues) rather than pine nuts. *P. armandii* Franch was often presented in the literature as the most notorious species connected to PNS. This species grows in and comes directly from China.

It is very important to understand the true value of forest products so that they are utilized sustainably and, if necessary, protected for harnessing future benefits. Most of the studies found during the course of the present review were associated to PNS. Fortunately, we found some literature related to the economic valuation of this nutritious product.

Masiero et al. [52], grouped 21 countries of the Mediterranean region into four sub-regions. This arbitrary grouping was done to better estimate values for a selected set of products including wood and non-wood forest products. Secondary data sources such as national reports, the FAO forest resource assessment report published in 2010, and seven additional papers were used to derive pine nuts results [52] (pp. 4–10). The pine nuts considered by Masiero et al. [52] were limited to *P. pinea*. The minimum and maximum economic values for the different product ranges of pine nuts were estimated. The two alternative scenarios were developed from the International Nut and Dried Fruit Council's report (min scenario) and the Mutke et al. (max scenario) paper published in 2012. The values estimated were 83.8 M€ to 307.7 M€ for producing 5295 tons to 18,992 tons of pine nuts. According to Masiero et al. [52], FAO Forest Resource Assessment (2010) indicated a market value of 48.7 M€ for the production of 16,545 tons of shelled pine nuts. Lebanon and Tunisia were regarded as important producers with respect to the minimum production scenario and Spain performed best when the maximum production scenario was applied.

Unfortunately, the Masiero et al. [52] study does not provide a holistic picture of different pine nut species. This is due to its specific focus on Mediterranean pine nuts. However, limitations due to a lack of official statistics and informal market channels for pine nuts justify the study.

Another study by Mutke et al. [27] also addressed and emphasized the importance of Mediterranean stone pines (*P. pinea*). A forest owner can obtain higher profits from pine nuts than harvesting timber in the Mediterranean region. In contrast to timber harvesting, revenue can be collected annually by selling pine kernels without waiting for the long pine rotations. In semi-arid sandy and/or rocky Mediterranean areas, the mean annual production of cones ranges 100–1000 kg ha^{−1} which can potentially yield 4–40 kg shelled pine nuts. So, prices in the world market are high enough (50–60 € kg^{−1}) to return an average income of 200–2400 € ha^{−1}.

A number of challenges along the European nuts' supply chain were of special interest in the paper by Mutke et al. [27]. It was noted that legal and commercial differentiation of those available on the market was without clear declaration of origin and species but under the generic name 'pine nuts'. For instance, in Germany *P. koraiensis* (Chinese nut) and *P. gerardiana* (Pakistani nut) were sold under the name *Pinienkerne* (general name for pine nuts in the German language). Similar issues were raised by Ballin and Mikklesen [42], who found *P. armandii* packed together with many other pine nut species.

Table 5. Summary of PNS studies.

Technique Used	Cases Reported	Case Study	Study Scope	References
Polymerase chain reaction (PCR) amplicon with principal component analysis (PCA)	NA	GC	SA	Ballin and Mikkelsen (2016) [42]
Used Taqman-based real time PCR method	NA	Italy (SC)	SA	Garino et al. (2016) [38]
Summarized the anaphylactic cases in the paper	45 cases	GC	RA	Cabanillas and Novak (2015) [26]
Genetic profiling of pine nuts	501 cases	USA (SC)	SA	Kwegyir-Afful et al. (2013) [47]
Gas chromatographic method	4 cases	Denmark (SC)	SA	Ballin (2012) [53]
Diagnostic index (DI) method and recently developed method in their laboratory	?	The USA (SC)	SA	Fardin-Kia et al. (2012) [37]
Descriptive analysis of cases	3111 cases	France (SC)	SA	Flesch et al. (2011) [45]
NA	1 case	The UK (SC)	L	Hampton et al. (2011) [46]
Genetic method	15 cases	USA (SC)	SA	Handy et al. (2011) [18]
Non-targeted nuclear magnetic resonance (NMR) spectroscopy and chemometrics	?	GC	SA	Köbler et al. (2011) [50]
Online survey form	107 cases	The Netherlands (SC)	GL	Tan (2011) [1]
NA	NA	The USA (SC)	GL	FDA (2011) [54]
NA	99% cases without referring to total number	The UK (SC)	GL	UKFSA (2011) [55]
Chemical and morphological analysis	NA	France (SC)	GL	ANSES (2010) [13]
Gas-liquid chromatography analysis	?	GC	SA	Destailats et al. (2010) [12]
NA	NA	Switzerland (SC)	L	Picard and Landis (2010) [44]
Standardized questionnaires as well as lab tests	800 cases	France (SC)	GL	AFSSA (2009) [56]
NA	?	Ireland (SC)	GL	FSAI (2009) [57]

Abbreviations used: GC = General case; GL = Grey; L = Letter to scientific journal; NA = Not applicable; SA = Scientific article; SC = Specific case; RA = Review article; ? = unknown.

The Sandeman Seeds Company labelled the UK as originator of those packets. However, we know that *P. armandii* nuts grow and come mainly from China. Mutke et al. [27] attributed confusion in detecting geographical origin of pine nuts to the concept of economies of scale (efficiencies formed by volume) rather than economies of scope (efficiencies formed by variety). This is due to a number of reasons such as the small size of pine nuts, dependence on a single product and supply, and current commercial and market structure. In our opinion, these issues most probably just happened due to re-exporting and re-importing of pine kernels.

The advent of modern device technology (mobiles and computers), internet, social networks and online news articles debating PNS have started to negatively affect trade and market development of pine nuts [27]. Although the international collection of food standards ‘Codex Alimentarius’ has categorized pine nuts in TN 0673 since 1993 [8], a clear system is still lacking for classifying the most important commercial species of pine nuts.

In some parts of the world, pine trees are mainly intended to better meet local consumption [8]. However, pine nuts might cause problems out of their native region. For instance, pine nuts imported in the US are the main cause of PNS establishment because of their mixed nature [39]. This is because pine nuts are difficult to distinguish morphologically. *P. gerardiana* is very similar to *P. pinea*. Similarly, *P. sibirica* resembles *P. armandii*. Ultimately, re-exports among several countries mask the traceability of origin and quality of the product. This is especially true when foreign suppliers normally sort and grade (from company A to C instead of B) pine kernels based on the number of pine nuts per 100 g. Business

as usual varies greatly based on seed sizes e.g., 650–750 nuts/100 g to 1500–1700 nuts/100 g [58]. This is why when pine nuts are purchased from a local market, species and geographical origin information is seldom on the packets.

As mentioned earlier, the number of reported cases of PNS started to increase in 2009. We have identified the following reasons in both the scientific as well as grey literature that might be directly or indirectly responsible. Contributing factors may be a combination of the following. (1) Pine nuts are extensively consumed worldwide as an ingredient in different foods, in both raw and roasted form. The growing popularity and introduction of pine nuts in dishes such as tarts, pesto and gourmet salads, desserts, breads, cakes, candies, vegetarian and non-vegetarian dishes, chocolates in Europe might be a contributing element [10,12,17–19,26,37,38,53]; (2) A rapid exchange of information by consumers via internet and the print media allow people to easily recognize the developing symptoms of PNS [17]; (3) Introduction of newer (non-traditional) species of pine nuts onto the global markets [17] as a result of competitive global trade.

The first two causes of PNS development might have given rise to the fact that many studies on pine nuts started to be done from the clinical toxicology, food and agricultural point of view. The third cause (i.e., global trade) was neglected as is evident from the dearth of studies. In our opinion, worldwide trade and the growing popularity of pine nuts in the food industry is a good opportunity for forest scientists, pathologists, and societal marketing researchers to take the initiative. The starting questions could be; which species would be good alternatives to those we already have? Which species are the most profitable? How can plantations be managed to obtain a bumper pine nuts crop? Why do a few tree species cause PNS while others do not?

Having said this, we acknowledge the fact that forest sciences research (particularly for societal marketing researchers and economists) on pine nuts might not be simple due to the lack of reliable data or a databank, which would hinder a full exploration of the trade aspects and valuation of pine nuts. However, it will be a challenge for forest pathologists and entomologists due to climate change and the increase in global trade of pine nuts, which are the main drivers of adventive insects. Table 5 summarizes the studies linked to PNS and economic valuations.

4.2.2. Pathogens and Pests of Pine Nuts

The introduction of emerging infectious diseases (EIDs) into new areas has been associated to climate change, human ecology, increased global trade and economic growth [59,60]. A recent study attributed seven underlying drivers (alien pathogenic invasion, climate change, emergence of aggressive species or strains, rise in hybridization of fungi, latent and cryptic pathogens, establishment of novel links between pathogens and their vectors, adaptation of new crops and cultivation practices) of EIDs [61]. Often more than one driver may cause a rise in the range of pathogens, insects and ultimately infectious diseases.

An increasing number of connections between adventive insects (acting as vector) and local fungi (acting as pathogen) occur in forest ecosystems [62,63]. According to Ghelardini et al. [61], establishment of novel links between pathogens and their vectors, climate change, latent and cryptic pathogens are a few of the important factors identified that cause tip-blight of pines, also called dieback, due to *Diplodia sapinea* or *D. pinea* and its association with an adventive insect, *Leptoglossus occidentalis* Heidemann. Figure 2 shows the current presence of *L. occidentalis* in the world. There are many cone and seed insects of pines mentioned in the literature. For example, *Dioryctria mendacella* Staudinger, *Ernobius parens* Mulsant and Rey, *Ernobius impressithorax* Pic, *Pissodes validirostris* Sahlberg, and *L. occidentalis* [64–68]. However, *L. occidentalis* is one of the most extensively studied insect vectors on pine nuts.

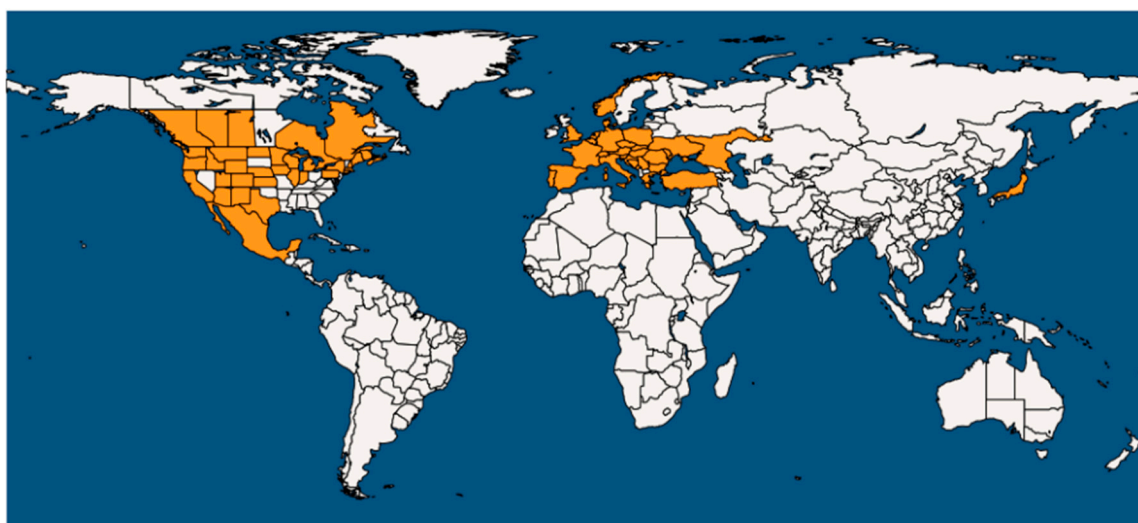


Figure 2. Current distribution of *Leptoglossus occidentalis* around the world (source: EPPO <http://www.eppo.int>).

D. sapinea is a widespread pathogenic fungus also known as *Sphaeropsis sapinea* or *D. pinea*. *D. sapinea* is commonly found in Europe, South Africa and the US. It is damaging for pines both in their natural range and in plantations [69–71]. *D. sapinea* is badly damaging the cones of stone pine (*P. pinea*) in Italy [72]. In 2013, the first report of *D. sapinea* on Scots pine (*Pinus sylvestris* L.) and Austrian pine (*Pinus nigra* (Lodd. ex Lindl.) Munro) came from Sweden [73]. The conidia of *D. sapinea* need *L. occidentalis* for their lengthy and extensive dispersal, though primary dissemination occurs via rain splashes and wind currents [74].

L. occidentalis (Hemiptera; Heteroptera; Coreidae) is an indigenous species of the US [75] and is commonly known as leaf-footed conifer seed bug or western conifer seed bug (WCSB). WCSB feeds on seed sap by sucking and weakens the seed or even makes it abort [76].

Among numerous introduced groups of organisms in Europe, arthropods represent nearly 94% [77]. WCSB is one of the insect species that was introduced into Europe roughly 20 years ago. It has been documented that it feeds on nearly 14 American and European conifer species: *Pinus* spp., *Calocedrus decurrens* (Torr.) Florin, *Pseudotsuga menziesii* (Mirb.) Franco, *Tsuga canadensis* (L.) Carrière, *Cedrus* spp., *Abies* spp. and *Picea* spp. [78,79] are some of the important ones. WCSB was observed for the first time in western North America. After World War II, it started to spread eastwards in the US and Canada [80–86]. It was recorded for the first time in Vicenza, Italy during autumn 1999 [75,87].

Soon after its discovery in Italy, WCSB started to expand to other parts of the European continent. It can now be found in Slovenia, Croatia, Hungary, Switzerland, Austria, Germany, and Czech Republic to the north, and France to the west [88]. In 2003, it was also independently recorded in Spain [88]. WCSB was also found for the first time in Japan in 2009 [89]. During 2010, it was reported in Denmark and Norway [90]. WCSB was first recorded in Russia in 2010 (Rostov Province) and in Ukraine in 2011 (Crimea and Zaporizhia Province) as cited by Gapon [91]. It was also first reported in Greece during 2011. The insect was collected from central Evia, Attica and the north of Peloponnese. It was mainly found on *Pinus halepensis* Miller [92]. Recently, some researchers confirmed the presence of WCSB in four regions of Chile i.e., Atacama, Metropolitan, Maule, and Bío Bío territories [93].

In northern Italy, WCSB can complete two or three generations per year [94]. In autumn, adults seek shelter for their overwintering activities [78]. The adults and nymphs feed on cones and can potentially create significant economic losses in high-value seed orchards [65,76,95]. WCSB has also been thought to deteriorate the production of pine nuts for human consumption in Italian stone pine (*P. pinea*) stands. Pine nuts production is decreased by the activities of WCSB in Italy, resulting in lower income [96] for forest owners. Furthermore, it is notorious for causing a nuisance and damage to plumbing materials

when adult aggregates invade residential buildings for overwintering [84,86,97]. This is why they are also known as urban bugs.

It is pertinent to say that beetles are usually associated as vector to plant pathogens in forests, however, this was the first time a true bug (*L. occidentalis*) was observed to play a possible role as vector for *D. sapinea* in a natural forest ecosystem [98]. During 2012, Luchi et al. [98] conducted an experiment to understand the fungus and disease outbreak mechanism. They found a correlation between the adventive *L. occidentalis* and native *D. sapinea* (*S. sapinea*). Molecular analysis (real-time PCR) showed that *D. sapinea* was present both on the bodies of *L. occidentalis* and on the symptomless *P. pinea* cones that were the insect's main food source. Moreover, both organisms share similar habitat conditions. The conspicuous damage on pine trees may be explained by their co-habitation that prompts the disease spread and great invasive potential [99].

The results of a research conducted by Bracalini et al. [68] at Maremma regional park in Italy also suggested that the low cone production and high number of damaged seeds agree with an overall decline of stone pine nut production in Tuscany (Italy). The authors claim that the findings have been confirmed by the local nut producing companies [68].

A fungus-insect interaction usually profits both parties [98]. The fungus takes advantage of the insect via its dispersal to other pines or other tree species while the insect profits because the fungus may stimulate the plant to produce monoterpenes, which are responsible for entomophilic community formation. Similarly, a study on *P. tabuliformis* (Chinese pine) reported that the fungus *Leptographium procerum* (W.B. Kendr.) (syn. *Verticicladiella proceri*) raised levels of monoterpene attracting the insect *Dendroctonus valens* L. [100] which acts as vector of the fungus.

Depending on climatic and environmental conditions, *D. sapinea* may act as a latent pathogen enhanced by climatic stress and able to cause a speedy death of currently expanding shoots [101]. The potential involvement of WCSB in vectoring the conidia of *D. sapinea* was verified by Tamburini et al. [87]. At present, *D. sapinea* and *L. occidentalis* are not listed as pests recommended for regulation of quarantine pests by the European and Mediterranean Plant Protection Organization [102].

5. Discussion and Conclusions

Information and data related to pine nuts are very limited in the literature. Most of the research is focused on phytosanitary problems affecting pines and the related nut production and on taste disturbance caused by pine nuts, PNS syndrome. The two issues are interlinked by the supply chain development: an increased demand for pine nuts has stimulated the development of international trade: to satisfy this increasing demand some pine nut species that cause PNS have been introduced in the market. Unfortunately, there is very limited work on the analysis of market development for these NWFP, partly due to recent growing demand in the western world and its informal market nature in some large producing countries. However, in this rapidly developing market a systematic organization, production presentation, and trade information by some global statistical institutions (e.g., FAOSTAT and UN COMTRADE), is required. In fact, we have found that pine nuts information in relation to its production, export and import is present in some regional statistical sources such as EUROSTAT, but this information is limited, with special emphasis on EU markets. There are inconsistencies of declared annual production and export data due to different levels of aggregation of the nut category or the use of generic names for the groups of various species.

As for any other food commodity affecting human health, there is a need to track pine nut species along the supply chain to prevent the introduction of invasive fungal and insect species. Ultimately, pine nut species need to be supported in terms of the management of pine forests that may have a relevant role in the economy and cultural value (landscape) of many countries, such as *P. pinea* in the Mediterranean region. Future research should focus more on the economic importance of this product, linking the pathological and entomological aspects of forest management to the human health aspects with the aim of consolidating a responsible supply chain organization. To increase the income generation from pine nuts more studies should be performed on integrated management of WCSB

(*L. occidentalis*) and *D. sapinea*, in addition to *D. mendacella*, *E. parens*, *E. impressithorax*, *P. validirostris*. Due to the billion-euro pine nuts industry all around world, legislative bodies should pay special attention to quarantine control of the wood movements containing potential pathogens and pests of pine nuts.

Supplementary Materials: The following are available online at <http://www.mdpi.com/1999-4907/8/10/367/s1>, Table S1.

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